Citation:

Hodge AM, English DR, O'Dea K, Sinclair AJ, Makrides M, Gibson RA, Giles GG. Plasma phospholipid and dietary fatty acids as predictors of type 2 diabetes: Interpreting the role of linoleic acid. *Am J Clin Nutr.* 2007 Jul; 86 (1): 189-197.

PubMed ID: 17616780

Study Design:

Prospective Cohort Study

Class:

B - Click here for explanation of classification scheme.

Research Design and Implementation Rating:

NEUTRAL: See Research Design and Implementation Criteria Checklist below.

Research Purpose:

To investigate the relationship between plasma phospholipid levels and dietary fatty acid intake and diabetes risk.

Inclusion Criteria:

- Subjects from the Melbourne Collaborative Cohort Study (MCCS) who were 40 to 69 years of age
- Individuals with incident cases of diabetes at follow-up and a random sample of the cohort without diabetes at follow-up.

Exclusion Criteria:

- Participants with diabetes at baseline
- Participants who had had a heart attack or had angina before baseline
- Those who did not report diabetes at baseline, but later reported a date of diabetes diagnosis before baseline
- Those with extreme self-reported energy intake (less than first percentile and higher than 99th percentile)
- Those with missing values for relevant risk factors.

Description of Study Protocol:

Design

Prospective case-cohort study.

Dietary Intake/Dietary Assessment Methodology

Self-administered 121-item food-frequency questionnaire (FFQ) specifically developed for MCCS

Statistical Analysis

- Means and SDs for each fatty acid in plasma phospholipid and diet were calculated by diabetes status at follow-up, and T-tests were used to evaluated differences between the two groups
- Age, country of birth, sex, physical activity score, five-year weight change, education level, smoking, BMI, waist-to-hip ratio and family history of diabetes were considered as potential confounders. Weight change, education and smoking were not associated with diabetes in the subcohort and were not included in subsequent models
- Logistic regression models were used to determine the relationship between quintiles or plasma phospholipid fatty acid proportions and dietary fatty acids expressed as energy density.

Data Collection Summary:

Timing of Measurements

- Baseline measures of diet and serum phospholipid levels were taken between 1990 and 1994
- Follow-up assessment of diabetes status was done approximately four years later.

Dependent Variables

- Diabetes diagnosis during follow-up was determined using a mailed self-administered questionnaire
- The authors attempted to verify with the person's doctor any reports of diabetes diagnosis
- Responses were available for 292 people and 291 were confirmed to have type 2 diabetes
- For the 52 people with no response and for the two people for whom doctors did not know the diabetes type, the authors assumed a diagnosis of type 2 diabetes.

Independent Variables

- Plasma phospholipid levels was assessed using fasting blood analysis
- Dietary intake of fatty acids was assessed using a 121-item FFQ
- The following fatty acids and classes were analyzed. Total saturated fatty acids (SFAs) were 15:0, 16:0, 18:0; total monounsaturated fatty acids (MUFAs) were 16:1n-7, 18:1n-9; total polyunsaturated fatty acids (PUFAs), total n-6 fatty acids, 18:2n-6, 20:3n-6, 20:4n-6; total n-3 fatty acids, 18:3n-3, 20:5n-3, 22:5n-3, 22:6n-3; ratio of n-6 to n-3 fatty acids, total trans fatty acids and total conjugated linoleic acid.

Control Variables

- Age
- Country of birth
- Sex
- Physical activity score
- Five-year weight change
- Education level
- Smoking
- BMI
- Waist-to-hip ratio

- Family history of diabetes.

Description of Actual Data Sample:

- *Initial N:* 41,528 subjects were recruited to participate in MCCS between 1990 and 1994
- *Attrition (final N):* 3,737 participants (2,088 women and 1,649 men), including 364 incident cases of type 2 diabetes, had complete data for these analyses
- *Age:*
  - Controls 54.5±8.6 years of age
  - Cases 57.9±7.3 years of age
- *Other relevant demographics:*
  - 17.6% of controls and 34.1% of cases had a family history of diabetes
  - 15.6% of controls and 34.1% of cases had primary education only
  - 24.4% of controls and 13.9% of cases were in the highest of four groups for physical activity score
- *Anthropometrics:*
  - Controls had a mean BMI of 26.5±4.2kg/m^2
  - Cases had a mean BMI of 31.7±5.2kg/m^2
- *Location:* Australia.

Summary of Results:

Plasma Phospholipids and Diabetes Incidence

- Persons who developed diabetes had higher plasma proportions of 18:0; total SFAs, 16:1n-7, 20:3n-6, 20:4n-6; total n-3 fatty acids, 20:5n-3 and 22:6n-3, and lower plasma proportions of of 15:0; total PUFAs, n-6 fatty acids, 18:2n-6, n-6:n-3, trans fats and conjugated linoleic acid at baseline than did persons who did not develop diabetes
- After adjustment for age, sex, country of birth, physical activity, family history of diabetes and alcohol intake, inverse associations were seen for 15:0, trans fatty acids, and 18:2n-6. Positive associations were observed for 18:0, total SFAs, 16:1n-7 and 20:3n-6.

Dietary Fatty Acids Intake and Diabetes Incidence

Persons who developed diabetes had higher plasma proportions of total fat, total MUFAs, 16:1n-7, 18:1n-9; total PUFAs, n-6 fatty acids, 18:2n-6, 20:4n-6, n-3 fatty acids, 18:3n-3, and trans fats, and lower intake of 15:0 at baseline than did persons that did not develop diabetes.

Fatty Acid	Controls (g per day)	Cases (g per day)	P-value
Total fat	82.16±30.52	86.25±34.22	0.033
SFAs	31.38±12.31	31.94±0.14	0.457
15:0	0.27±0.14	0.25±0.14	0.041
16:0	16.28±6.04	16.96±6.78	0.071
18:0	7.79±3.14	8.06±3.50	0.160
MUFAs	28.67±10.76	31.60±12.99	0.0001
16:1n-7	1.75±0.69	1.89±0.80	0.001

18:1n-9	26.28±9.94	29.03±12.11	0.0001
PUFAs	12.60±5.31	13.62±6.86	0.007
Total n-6	11.34±4.99	12.28±6.47	0.009
18:2n-6	11.28±4.98	12.20±6.46	0.010
20:3n-6	0.02±0.02	0.03±0.03	0.078
10:4n-6	0.041±0.03	0.045±0.03	0.018
Total n-3	1.25±0.48	1.35±0.53	0.002
18:3n-3	0.95±0.33	1.03±0.44	0.0006
20:5n-3	0.10±0.09	0.10±0.07	0.568
22:5n-3	0.03±0.03	0.03±0.03	0.587
22:6n-3	0.18±0.15	0.18±0.11	0.509
n-6:n-3	9.17±2.81	9.16±2.83	0.935
Total trans	0.10±0.11	0.12±0.13	0.004

- The top quintile of dietary fat intake had an elevated risk of diabetes compared to the lowest quintile (1.59, 95% CI 1.08, 2.33)
- Both 16:0 (1.65, 95% CI 1.12, 2.43) and 18:0 (1.46, 95% CI 1.00, 2.14), but not total SFAs, were associated with higher risk of diabetes
- 16:1n-7 showed a weak positive association with diabetes (1.33, 95% CI 0.92-1.90)
- n-6:n-3 showed a positive association with diabetes (1.56 (95% CI 1.03, 2.36)
- Positive associations were seen for 18:1n-9, MUFAs, 18:2n-6, total n-6, PUFAs and 18:3n-3, but after adjustment for body size, the associations were no longer significant.

Other Findings

- Within each quintile of reported dietary intake, persons who developed diabetes had lower mean plasma phospholipid linoleic acid proportions, 1.8 (95% CI 1.4, 2.1)
- There was a weak interaction between plasma insulin and linoleic acid intake (P=0.09), and the association between dietary linoleic acid and diabetes risk was most apparent in persons with plasma insulin concentrations at or above the median value (5.3pmol per L or more). The OR for quintile five vs. quintile one was 1.81 (95% CI 1.01, 3.23, P=0.02).
- There was no significant difference in the associations of dietary linoleic acid with incident diabetes across strata of age or BMI.

Author Conclusion:

- This study found positive associations between the incidence of diabetes and SFAs in plasma phospholipid and diet
- Plasma linoleic acid was inversely, and dietary linoleic acid was positively, associated with diabetes risk
- Persons who developed diabetes had lower plasma phospholipid linoleic acid proportions for each quintile of linoleic acid intake than did persons with diabetes.

Reviewer Comments:

None.

Research Design and Implementation Criteria Checklist: Primary Research

Relevance Questions		
1.	Would implementing the studied intervention or procedure (if found successful) result in improved outcomes for the patients/clients/population group? (Not Applicable for some epidemiological studies)	Yes
2.	Did the authors study an outcome (dependent variable) or topic that the patients/clients/population group would care about?	Yes
3.	Is the focus of the intervention or procedure (independent variable) or topic of study a common issue of concern to nutrition or dietetics practice?	Yes
4.	Is the intervention or procedure feasible? (NA for some epidemiological studies)	Yes

Validity Questions		
1.	**Was the research question clearly stated?**	Yes
1.1.	Was (were) the specific intervention(s) or procedure(s) [independent variable(s)] identified?	Yes
1.2.	Was (were) the outcome(s) [dependent variable(s)] clearly indicated?	Yes
1.3.	Were the target population and setting specified?	Yes
2.	**Was the selection of study subjects/patients free from bias?**	No
2.1.	Were inclusion/exclusion criteria specified (e.g., risk, point in disease progression, diagnostic or prognosis criteria), and with sufficient detail and without omitting criteria critical to the study?	Yes
2.2.	Were criteria applied equally to all study groups?	Yes
2.3.	Were health, demographics, and other characteristics of subjects described?	Yes
2.4.	Were the subjects/patients a representative sample of the relevant population?	No
3.	**Were study groups comparable?**	Yes
3.1.	Was the method of assigning subjects/patients to groups described and unbiased? (Method of randomization identified if RCT)	N/A
3.2.	Were distribution of disease status, prognostic factors, and other factors (e.g., demographics) similar across study groups at baseline?	N/A

3.3.	Were concurrent controls used? (Concurrent preferred over historical controls.)	Yes
3.4.	If cohort study or cross-sectional study, were groups comparable on important confounding factors and/or were preexisting differences accounted for by using appropriate adjustments in statistical analysis?	Yes
3.5.	If case control or cross-sectional study, were potential confounding factors comparable for cases and controls? (If case series or trial with subjects serving as own control, this criterion is not applicable. Criterion may not be applicable in some cross-sectional studies.)	Yes
3.6.	If diagnostic test, was there an independent blind comparison with an appropriate reference standard (e.g., "gold standard")?	N/A
4.	**Was method of handling withdrawals described?**	Yes
4.1.	Were follow-up methods described and the same for all groups?	Yes
4.2.	Was the number, characteristics of withdrawals (i.e., dropouts, lost to follow up, attrition rate) and/or response rate (cross-sectional studies) described for each group? (Follow up goal for a strong study is 80%.)	Yes
4.3.	Were all enrolled subjects/patients (in the original sample) accounted for?	Yes
4.4.	Were reasons for withdrawals similar across groups?	???
4.5.	If diagnostic test, was decision to perform reference test not dependent on results of test under study?	N/A
5.	**Was blinding used to prevent introduction of bias?**	Yes
5.1.	In intervention study, were subjects, clinicians/practitioners, and investigators blinded to treatment group, as appropriate?	N/A
5.2.	Were data collectors blinded for outcomes assessment? (If outcome is measured using an objective test, such as a lab value, this criterion is assumed to be met.)	N/A
5.3.	In cohort study or cross-sectional study, were measurements of outcomes and risk factors blinded?	Yes
5.4.	In case control study, was case definition explicit and case ascertainment not influenced by exposure status?	N/A
5.5.	In diagnostic study, were test results blinded to patient history and other test results?	N/A
6.	**Were intervention/therapeutic regimens/exposure factor or procedure and any comparison(s) described in detail? Were interveningfactors described?**	Yes
6.1.	In RCT or other intervention trial, were protocols described for all regimens studied?	N/A

6.2.	In observational study, were interventions, study settings, and clinicians/provider described?	Yes
6.3.	Was the intensity and duration of the intervention or exposure factor sufficient to produce a meaningful effect?	Yes
6.4.	Was the amount of exposure and, if relevant, subject/patient compliance measured?	Yes
6.5.	Were co-interventions (e.g., ancillary treatments, other therapies) described?	N/A
6.6.	Were extra or unplanned treatments described?	N/A
6.7.	Was the information for 6.4, 6.5, and 6.6 assessed the same way for all groups?	Yes
6.8.	In diagnostic study, were details of test administration and replication sufficient?	N/A
7.	**Were outcomes clearly defined and the measurements valid and reliable?**	Yes
7.1.	Were primary and secondary endpoints described and relevant to the question?	Yes
7.2.	Were nutrition measures appropriate to question and outcomes of concern?	Yes
7.3.	Was the period of follow-up long enough for important outcome(s) to occur?	Yes
7.4.	Were the observations and measurements based on standard, valid, and reliable data collection instruments/tests/procedures?	Yes
7.5.	Was the measurement of effect at an appropriate level of precision?	???
7.6.	Were other factors accounted for (measured) that could affect outcomes?	Yes
7.7.	Were the measurements conducted consistently across groups?	Yes
8.	**Was the statistical analysis appropriate for the study design and type of outcome indicators?**	Yes
8.1.	Were statistical analyses adequately described and the results reported appropriately?	Yes
8.2.	Were correct statistical tests used and assumptions of test not violated?	Yes
8.3.	Were statistics reported with levels of significance and/or confidence intervals?	Yes
8.4.	Was "intent to treat" analysis of outcomes done (and as appropriate, was there an analysis of outcomes for those maximally exposed or a dose-response analysis)?	No
8.5.	Were adequate adjustments made for effects of confounding factors that might have affected the outcomes (e.g., multivariate analyses)?	Yes

8.6.	Was clinical significance as well as statistical significance reported?	Yes
8.7.	If negative findings, was a power calculation reported to address type 2 error?	No
9.	**Are conclusions supported by results with biases and limitations taken into consideration?**	Yes
9.1.	Is there a discussion of findings?	Yes
9.2.	Are biases and study limitations identified and discussed?	No
10.	**Is bias due to study's funding or sponsorship unlikely?**	Yes
10.1.	Were sources of funding and investigators' affiliations described?	Yes
10.2.	Was the study free from apparent conflict of interest?	Yes